PATENT SPECIFICATION

NO DRAWINGS

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Inventors:—JAMES GROVENOR BAXTER and ORRIS DURAND HAWKS.

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COMPLETE SPECIFICATION

Treatment of Xanthophyll Concentrate

We, EASTMAN KODAK COMPANY, a Company organized under the Laws of the State of New Jersey, United States of America of 343 State Street, Rochester, New York 14650, United States of America, do hereby declare the invention, for which we pray that a patent may be granted to us, and the method by which it is to be performed, to be particularly described in and by the following statement:—

This invention relates to xanthophyll.

Xanthophyll is a known generic term for certain oxygenated carotenoids found in many plants. As used herein, the term xanthophyll is intended to include one or a mixture of such compounds. Xanthophyll has been found to be useful as a yellow colouring agent for various foodstuffs for human consumption, and has been added to poultry feed so as to develop a yellow skin and shank pigmentation in the poultry, which is sometimes considered to be desirable. The yolk of eggs laid by the poultry have a strong yellow colouration, and such eggs are in demand for the manufacture of various foodstuffs such as egg noodles and yellow cake.

The pigmenting activity of xanthophyll varies according to the source from which it is obtained. A correlation has been observed between the pigmenting activity of a material and what is known as its alkali consumption value.

The alkali consumption value of a material is the number of milligrams of potassium hydroxide consumed by chemical reaction when 1 gram of the material is admixed with potassium hydroxide in 95% ethanol, and established and maintained at one hour at boiling point temperature, the quantity potassium hydroxide in 95% ethanol being selected so that on back titration with 0.5 N hydrochloric acid, with phenolphthalein as an indicator, the volume of acid used is 45-55% of the volume of acid used in titrating the quantity of potassium hydroxide in 95% ethanol in the absence of the material.

It has been observed that, in general, xanthophyll having a substantial alkali consumption value has less than maximum pigmenting activity.

In our British Patent Specification No. 1,046,658 it has been proposed to treat such xanthophyll with sodium or potassium hydroxide, preferably in a quantity at least chemically equivalent to the alkali consumption value and at a temperature in the range from 0 to 150°C. Such treatment increases the pigmenting activity of the xanthopyhll.

The xanthophyll to be treated according to this invention is usually that found in non-leafy plant material, but any leafy plant material which contained xanthophyll having a substantial alkali consumption value could also be used.

A useful source of xanthophyll having a substantial alkali consumption value is the petals of the flower of the Aztec marigold (Tagetes erecta L.).

Dried and powdered whole marigold flowers and petals, generically called marigold meal, have been proposed for addition to poultry feed, but the pigmenting activity of the xanthophyll contained therein is not as great as desired. A xanthophyll concentrate having a low or no alkali consumption value may be prepared by methods disclosed in our afore-mentioned British Patent Specification, and by other methods. The percentage of xanthophyll in such a concentrate tends to diminish with the passage of time, probably due to atmospheric oxidation, and the addition of at least one antioxidant such as ethoxyquin (6-ethoxy-2,2,4-trimethyl-1,2-dihydroquinoline) or toco-

[Price 4s. 6d.]

pherol may increase the stability of the concentrate.

It is an object of the invention to provide a method of treating a xanthophyll concentrate so as to improve its stability.

The invention consists in a method of treating a xanthophyll concentrate prepared from vegetable matter containing xanthophyll, the concentrate containing at least one antioxidant for xanthophyll, comprising adding thereto at least one fat or fatty-like material, and heating so as to improve the stability of the xanthophyll concentrate.

Preferably the concentrate is marigold meal, and at least 5% by weight, based on the weight of the marigold meal, of said at least one fat or fatty-like material is added.

It is not entirely clear why the addition of fat or fatty-like material has a beneficial effect, but it may be that it brings the xanthophyll into better contact with the antioxidant because of their mutual solubility in the fat.

The term "fat or fatty-like material" is intended to include both fats and oils, the free fatty acids or derived lipids, and glycerides or fatty alcohols. The fat may be an edible vegetable fat, such as soybean oil, cottonseed oil, corn oil, or sunflower seed oil. It may be of animal origin such as beef or pig tallow, or other tallows and greases. Whale oil and fish oils and fats generally can also be used.

While even small amounts of fat enhance the stability of the product, the most advantageous results are obtained with at least 5% of fat, based on the weight of the marigold meal; and an upper limit of 35% of fat is an economical one, since the increased addition of fat above this upper limit is not accompanied by a commensurate improvement in stability. The preferred range is 10 to 20%.

Heating is preferably carried out at from 70 to 180°C.

The time required for heat treatment diminishes as the temperature increases so that it is normally desirable to use higher temperatures, such as 110°C. to 165°C. The operable upper limit is 180°C., but higher temperatures may tend to produce degradation of the xanthophyll. Substantially the same stabilization results are secured by heating for one minute or less at 165°C as by heating for ten hours at 70°C.

When the xanthophyll concentrate contains liquid, the heat treatment may be carried out conveniently and effectively by drum drying. For example, an aqueous slurry of alkali-treated marigold meal, containing antioxidants and fat, may be dried on a drum drier, with rolls at a selected temperature, e.g. at a temperature between 110°C. and 165°C., running at a speed of between one and four RPM. Spray drying has also been used successfully.

The xanthophyll concentrate may be prepared from vegetable matter by the steps of:

a) digesting a slurry of the vegetable matter in a mixture of an alkali metal hydroxide with an alkanol having up to 6 carbon atoms;
b) neutralizing the slurry thereby forming a salt therein;
c) adding sufficient water to dissolve the salt and any other water-soluble material present;
d) separating the aqueous solution thus formed from the water-insoluble portion of the digested vegetable matter; and
e) recovering the water-insoluble portion as the xanthophyll concentrate; said at least one fat or fatty-like material being added at any stage after step (b).

This procedure makes it possible to obtain a higher concentration of xanthophyll than that which is obtained by the procedures disclosed in our afore-mentioned British Patent Specification, because of the removal of the salt resulting from neutralization of the alkali, and the removal of any other water - soluble non - xanthophyll materials.

The xanthophyll concentrate prepared in this manner may have, for example, from two to ten times the xanthophyll concentration of the starting material, e.g. marigold meal. The concentrate prepared from marigold meal comprises solid marigold fibres carrying the xanthophyll.

The addition of fat or fatty-like material cannot be carried out before alkali treatment, or a soap would be formed. The material can be added at any stage after neutralization, as will be seen in the following Examples which are illustrative of the invention.

In the Examples, the xanthophyll concentration of a material is measured by measuring the absorption of light of wavelength 453 mμ by a sample 1 cm. thick containing 1% of the material in chloroform. This absorption is recorded as E(1%, 1 cm.) (453 mμ), and is 2400 for pure xanthophyll.

EXAMPLE 1

Marigold petal meal (200 g.), [E (1%, 1cm) (453 mμ)=33.1] was mixed with methanol (300 ml.) and sodium hydroxide (57 g.) dissolved in water (50 ml.) was added. The resulting slurry was kept under reflux for 2 hours and neutralized to pH 7.5 with 50% w/w aqueous phosphoric acid (about 76 g.). Ethoxyquin (0.45 g.) and tocopherol (0.45 g.) (antioxidants) and tallow (20 g.) were added. The alcohol was removed by distillation and replaced by addition of water. The aqueous slurry was filtered, after which the filter cake was

drum dried to give 80 g. of product having E (1%, 1cm) (453 mμ)=82.6. The aqueous filtrate contained no detectable amounts of xanthophyll.

A duplicate experiment, except that the washing and filtration steps were omitted, gave a product having E (1%, 1cm) (453 mμ)=20.6. Thus, the xanthophyll concentration in the water-washed and filtered product was four times that of the unwashed product.

EXAMPLE 2

Marigold petal meal (29.5 g.), [E (1%, 1cm) (453 mμ)=33.1] was mixed with ethanol (45 ml.) and treated with sodium hydroxide (8.4 g.) dissolved in water (7.5 ml.). The reaction mixture was refluxed for 2 hours, cooled, and neutralized to pH 7.5 with 15% aqueous phosphoric acid (about 37 g.). Tocopherol (70 mg.) and ethoxyquin (70 gm.) were added as antioxidants and the ethanol distilled off. The resulting slurry was added to cold water (600 ml.), and stirred and filtered. Molten tallow (3 g.) was mixed into the filter cake, which was then drum dried to give 13.9 g of a product having E (1%, 1cm) 453 mμ)=69. The xanthophyll concentration was 3.5 times as high as in the product of Example 1 which was processed without water washing.

EXAMPLE 3

Marigold petal meal (11.8 g.), [E (1%, 1cm) (453 mμ)=33.1] was mixed with ethanol (18 ml.) and treated with sodium hydroxide (3.4 g.) dissolved in water (3.0 ml). The reaction mixture was refluxed for 2 hours, cooled and neutralized to pH 7.5 with 15% w/w aqueous phosphoric acid (about 15 g.). Ethoxyquin (28 mg.) and tocophenol (28 mg.) were added as antioxidants and the aqueous alcohol slurry poured into water (200 ml.). The mixture was stirred for 1 hour and then filtered. Tallow (1.2 g.) was added to the filter cake, which was dried to give 5.6 g. of a product having E (1%, 1cm) (453 mμ)=63. The xanthophyll concentration was three times that of the unwashed product of Example 1.

EXAMPLE 4

Marigold whole flower meal (100 g.), [E (1%, 1cm) (454 mμ)=15.6] was mixed with ethanol (300 ml.) and treated with sodium hydroxide (29 g.) in water (50 ml.). The mixture was refluxed for 2 hours, cooled, and neutralized to pH 7.2 with 10% w/w aqueous phosphoric acid (about 190 g.). Tocopherol (0.112 g.) and ethoxyquin (0.225 g.) were added as antioxidants, the ethanol was boiled off and water (2000 ml.) added. The slurry was filtered, and tallow (5 g.) and stearic acid (2.5 g.) were added to the filter cake, which was then drum dried to give 52 g. of a product having E (1%, 1cm)(454 mμ)=26. The xanthophyll concentration was 2.5 times that found for a comparison sample of the unwashed product. [E (1%, 1cm) (454 mμ)= approximately 10].

EXAMPLE 5

Marigold whole flower meal (100 g.), [(1%, 1 cm)(454 mμ)=15.6] was mixed with ethanol (300 ml.) and treated with sodium hydroxide (29 g.) in water (50 ml.). The mixture was refluxed for 2 hours, cooled and neutralized to pH 7.2 with 10% w/w aqueous phosphoric acid (about 190 g.). Tocopherol (0.112 g.) and ethoxyquin (0.225 g.) were added, the ethanol was boiled off and water (2000 ml.) added. This slurry was centrifuged and the supernatant liquid decanted. Tallow (5 g.) and stearic acid (2.5 g.) were added to the centrifuged cake, which was drum dried to give 57.2 g. of a product having E (1%, 1cm) (454 mμ)= 25. The xanthophyll concentration was 2.5 times that found for a comparison sample of the unwashed product.

EXAMPLES 6 TO 9

Four portions, each of 200 grams, of marigold petal meal were each treated as in Example 1 up to the neutralization stage. Neutralization to pH 7.5 was effected with a different acid for each portion, as indicated below.

Example	*Acid*	*Amount in grams*
6	50% w/w aqueous HCl	28
7	50% w/w aqueous H_2SO_4	40
8	50% w/w aqueous H_2SO_3	32
9	acetic acid	24

Following neutralization, the rest of the treatment on each portion was performed as described in Example 1. The resulting water-washed and filtered products were found to be comparable in xanthophyll content to the water-washed and filtered product of Example 1.

EXAMPLE 10

200 grams of marigold meal [1.4% xanthophyll E (1%, 1cm)(454 mμ)=32, alkali consumption value=223] were slurried in 600 ml. of ethanol and 0.225 gram of ethoxyquin was added. 50 grams of sodium hydroxide dissolved in 35 ml. of hot water were added to the slurry, which was then heated at a temperature of about 80°C. under reflux conditions for two hours. The slurry was then neutralized with a 25% solution of phosphoric acid in water, and 0.225 gram of ethoxyquin and 0.45 gram of tocopherol were added as antioxidants.

The product was then divided into four fractions and different amounts of a mixture of two parts tallow to one part stearic acid by weight were added to three of the fractions and mixed intimately therein. With all four fractions, the ethanol was removed by distillation and was replaced with water until an essentially aqueous slurry was ob-

tained. Each fraction was then divided into two portions, A and B. Each portion A of aqueous slurry was spray dried in a spray drier having an inlet temperature of 260°C. and an outlet temperature of 70°C. Each portion B of slurry was drum dried on a drum heated with steam supplied at 140°C. to which it was subjected for about 30 seconds, the temperature when the dried product was removed from the drum being about 120°C.

The resulting dried products were analyzed for xanthophyll content, and were then placed in an oven at 43°C. and kept for one week, after which the products were again analyzed for xanthophyll content. The analysis involved extracting the products with chloroform and determining the xanthophyll content by measurement of absorption spectra. The results are shown in Table I.

TABLE I

Fraction	*Tallow : stearic acid %*	*% Xanthophyll Recovery Portion A: Spray Dried, Stored 1 week at 43°C.*	*% Xanthophyll Recovery Portion B: Drum Dried, Stored 1 week at 43°C.*
1	0	64.3	82.0
2	5	69.7	91.2
3	10	98.2	101.0
4	15	96.5	99.6

The figure of 101.0% obtained with Portion B of fraction 3 is, of course, greater than the theoretical maximum of 100%, but this is due to experimental error in the absorption measurements.

It will be observed from the foregoing data that the addition of fat combined with heat treatment increased the stability of the xanthophyll in both the drum dried product and the spray dried product. The greatest improvement of stability occurred with the combination of fat addition and the more rigorous heat treatment by drum drying. It is thought that the higher temperature of drum drying gives better contact between the xanthophyll, the fat and the antioxidants, and thus gives improved incapsulation. While the spray drier has a higher inlet temperature, the sprayed material is not treated at this temperature.

EXAMPLE 11

A neutralized slurry containing antioxidants was prepared by the procedure of Example 10.

The product was divided into four fractions, and 15% by weight of different fats were added to three of the fractions and mixed intimately therein. The four fractions were then treated by the procedure of Example 10, each fraction being divided into two portions, A and B.

The results of the determinations of xanthophyll content are shown in Table II.

TABLE II

Fraction	*Fat Added (15%)*	*% Xanthophyll Recovery Portion A: Spray Dried, Stored 1 week at 43°C.*	*% Xanthophyll Recovery Portion B: Drum Dried, Stored 1 week at 43°C.*
1	Tallow : stearic acid (2:1)	96.5	99.6
2	Stearic acid	Not Determined	96.1
3	Glycerol monostearate	97.1	101.0
4	None	64.3	82.0

Again, stability was improved by fat addition even in the spray dried product, but the combination of fat and drum drying gave the best results.

EXAMPLE 12

Following the procedure of Example 10, the effect of various fats in combination with drum drying, with the rolls at a temperature developed by steam at 140°C., was investigated, with the results shown in Table III:

TABLE III

Fraction	Fat	Wt. %	Storage time	% Xanthophyll Recovery
1	Yellow grease	15	1 week at 43°C.	87
2	Glycerol mono-oleate	10	12 days at 43°C.	97
3	Corn oil	10	6 months at room temp.	96
4	Acetylated glycerol monostearate	10	1 week at 43°C.	97
5	Tallow	15	8 days at 43°C.	100

EXAMPLE 13

3 kilograms of marigold petal meal were slurried in 7.5 kilograms of ethanol, and a solution of 0.63 kilogram of water was hydroxide in 0.4 kilogram of water was added. The resulting slurry was stirred and refluxed at a temperature of about 80°C. for 1.5 hours, and then neutralized to a pH of 8 with a solution of 2.5 kilograms of 86% phosphoric acid in ethanol (16.7%). 0.3 kilogram of tallow, 0.15 kilogram of stearic acid, 12 grams of ethoxyquin, and 24 grams of tocopherol were then added.

Ethanol was removed by distillation and released with water until an essentially aqueous slurry was obtained. Five fractions of this slurry were drum dried under different conditions, and the stability of the dried products was determined by storage for one week at 43°C. (Xanthophyll analysis was conducted before and after storage, as described in Example 10).

Table IV describes the conditions of the drying operation and records the xanthophyll recovery which was found.

TABLE IV

Drum Drier Settings

Fraction	steam p.s.i.	steam temp. °C.	rolls r.p.m.	gap. in.	% Xanthophyll Recovery
1	40	140	3.5	.008	87.4
2	40	140	1.75	.008	89.7
3	65	155	3.5	.008	91.0
4	65	155	1.75	.008	96.3
5	65	155	3.5	.004	94.8

It will be observed from Table IV that a more stable product is obtained with a more rigorous heat treatment.

EXAMPLE 14

Marigold petal meal (200 grams), [E(1%, 1cm) (453 mμ)=29.4] was saponified, refluxed, cooled and neutralized with aqueous phosphoric acid to a pH of 7.1 in a manner similar to that described in Examples 2-5. Ethoxyquin (0.3 gram per gram of xanthophyll content) and tocopherol (0.6 gram per gram of xanthophyll content) were added as antioxidants, the alcohol was boiled off and 20 volumes of water per unit volume of slurry were added. The slurry was centrifuged and the supernantant liquid was decanted. The resulting centrifuged cake was divided into three fractions, to one of which was added 10% of tallow and to another 10% of tallow and 5% of stearic acid, the percentages being by weight. The three cake fractions were then dried for two hours in an externally heated laboratory rotary evaporator having an internal temperature of about 70°C.

The results shown in Table V below illustrate the effect of storage on the end product for various periods of time at 20°C.

Table V

Fraction	Fat	Initial E Value	% Xanthophyll Recovery after number of weeks shown 1	2	4	8
1	none	62	100	94	72	64
2	10% tallow	56.9	98	95	80	81
3	10% tallow+ 5% stearic acid	52.8	100	96	95	96

EXAMPLE 15

The procedure described in Example 14 was repeated with the exception that neutralization was carried out to a pH of 7.1, 10% by weight of tallow and 5% by weight of stearic acid were incorporated in the whole of the centrifuged cake and the latter was drum dried using steam at about 155°C.

The results shown in Table VI below illustrate the effect of storage on the end product for different times and temperatures.

Table VI

% Xanthophyll Recovery

Initial E Value	*1 week at 43°C.*	*1 week at 20°C.*	*8 weeks at 20°C.*
40.7	96.5	99.3	93.2

WHAT WE CLAIM IS:—

1. A method of treating a xanthophyll concentrate prepared from vegetable matter containing xanthophyll, the concentrate containing at least one antioxidant for xanthophyll, comprising adding thereto at least one fat or fatty-like material, and heating so as to improve the stability of the xanthophyll concentrate.

2. A method as claimed in Claim 1, wherein the concentrate is marigold meal, and at least 5% by weight, based on the weight of the marigold meal, of said at least one fat or fatty-like material is added.

3. A method as claimed in Claim 2, wherein from 10 to 20% by weight, based on the weight of the marigold meal, of said at least one fat or fatty-like material is added.

4. A method as claimed in Claim 1, 2 or 3, wherein heating is carried out at a temperature of from 70 to 180°C. for a time sufficient to improve the stability of the xanthophyll concentrate.

5. A method as claimed in any one of Claims 1 to 4, wherein the xanthophyll concentrate contains liquid, and after the addition of said at least one fat or fatty-like material, is heated by spray drying so as to improve the stability of the xanthophyll concentrate.

6. A method as claimed in any one of Claims 1 to 4, wherein the xanthophyll concentrate contains liquid, and after the addition of said at least one fat or fatty-like material, is heated by drum drying so as to improve the stability of the xanthophyll concentrate.

7. A method as claimed in any one of Claims 1 to 6, wherein the xanthophyll concentrate is prepared from vegetable matter by the steps of:

a) digesting a slurry of the vegetable matter in a mixture of an alkali metal hydroxide with an alkanol having up to 6 carbon atoms;

b) neutralizing the slurry thereby forming a salt therein;

c) adding sufficient water to dissolve the salt and any other water-soluble material present;

d) separating the aqueous solution thus formed from the water-insoluble portion of the digested vegetable matter; and

e) recovering the water-insoluble portion as the xanthophyll concentrate; said at least one fat or fatty-like material being added at any stage after step (b).

8. A method as claimed in Claim 7, including the step of drying the water-insoluble portion.

9. A method as claimed in Claim 7, wherein the aqueous solution is separated from the water-insoluble portion by filtering or centrifuging.

10. A method as claimed in any one of Claims 7 to 9, wherein the lower alkanol is methanol or ethanol.

11. A method as claimed in any one of Claims 7 to 10, wherein the slurry is neutralized with phosphoric, sulphuric, sulphurous, acetic or hydrochloric acid.

12. A method as claimed in any one of Claims 7 to 11, wherein said at least one antioxidant is added to the slurry.

13. A method of treating a xanthophyll concentrate as claimed in Claim 1 and substantially as hereinbefore described in any one of the Examples.

14. A treated xanthophyll concentrate prepared by a method as claimed in any one of Claims 1 to 6.

15. A treated xanthophyll concentrate prepared by a method as claimed in any one of Claims 7 to 12.

16. A treated xanthophyll concentrate prepared by a method as claimed in Claim 13.

17. Poultry food containing a treated xanthophyll concentrate as claimed in Claim 14, 15 or 16.

L. A. TRANGMAR, B.Sc., C.P.A.,
Agent for the Applicants.

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